

## Hygiena<sup>®</sup> Prep Xpress Protocol Guide

The Prep Xpress automation instrument is designed for high-throughput liquid handling. These protocols have been optimized for high throughput utilizing the 8-well multi-channel pipette head. This instrument is complementary to the BAX<sup>®</sup> System sample preparation protocols for filling cluster tubes with lysis solution or BHI media, transferring from a sample or BHI cluster tube into lysis solution cluster tubes, or transferring from prepared lysate cluster tubes into PCR tubes. This instrument does not perform onboard heating steps. Therefore, the use of a manual or automated heating block is required to complete portions of the BAX System sample preparation protocols.

### Labware:

Labware should be installed prior to performing any protocols. Using an admin profile, navigate to Settings > Software > File Manage > Labware > Import (3-line menu) to import all labware and mark Hygiena-specific labware as favorites.

- MIS2103** - Plate: Hygiena Cluster Tube Rack w Adapter 96 Green 1.1 mL
- MIS2104** - Reservoir: Vblock 250 mL Open Reservoir, V-bottom, Clear PP
- MIS2111** - Reservoir: Seahorse-Agilent, Reservoir, 1 cavity, 8 Row, 290ml, Clear PP
- MIS2107** - Tips: 50 µL Framed, Conductive Filtered
- MIS2082** - Tips: 300 µL Framed, Conductive Filtered
- MIS2108** - Tips: 1,000 µL Framed, Conductive Filtered
- MIS2109** - Rack: Eppendorf PCR-Cooler, 200 µL, Strip-Well, PCR, White Well
- MIS2110** - Rack: Prep Xpress 5 mL Tube Rack

### Protocols:

Protocols should be installed and verified prior to use. Using an admin profile, navigate to Settings > Software > File Manage > Protocols > Import (3-line menu). Following import, ensure that **each** protocol has been verified by selecting **Edit**, then **Verify**. **Prior to initiating any runs**, ensure calibration has been performed within the month of installation of new protocols.

- Prep-Lysis\_Solution\_(6\_racks)
- Prep-BHI\_Solution\_(5\_Racks)
- Transfer-5\_uL\_Cluster\_Tube\_to\_Cluster\_Tube
- Transfer-20\_uL\_Cluster\_Tube\_to\_Cluster\_Tube
- Hydrate-30\_uL\_Lysate\_to\_PCR
- Hydrate-50\_uL\_Lysate\_to\_PCR
- Hydrate-30\_uL-Lysate\_to\_PCR\_SINGLE
- Hydrate-50\_uL-Lysate\_to\_PCR\_SINGLE
- Sample\_tubes\_to\_cluster\_tube\_rack
- Sample\_tubes\_to\_cluster\_tube\_rack-5\_uL
- Sample\_tubes\_to\_cluster\_tube\_rack-10\_uL
- Sample\_tubes\_to\_cluster\_tube\_rack-20\_uL

All protocols and labware must be installed by a Hygiena Technical Support Specialist. Please visit our [website to contact](#) us for any installation, training, or troubleshooting assistance.

## Prep-Lysis Solution (6 racks)

- 6 – Cluster Tube Rack Adapters
- 6 – Green Cluster Tube Racks w/Cluster Tubes
- 1 – Vblock 250 mL Open Reservoir
  - Combined Lysis Buffer & Protease Solution
- 1 – Rack of 300  $\mu$ L Framed Conductive Filter

**Capacity:** Up to 6 racks of 96 cluster tubes (576 samples)

**Tips Utilized:** 8 - 300  $\mu$ L tips

**Pipette Used:** 8-channel (no individual wells)



## Prep-BHI Solution (5 Racks)

- 5 – Cluster Tube Rack Adapters
- 5 – Green Cluster Tube Racks w/Cluster Tubes
- 1 – 290 mL Seahorse-Agilent Reservoir
  - BHI Media
- 1 – Rack of 1,000  $\mu$ L Framed Conductive Filter
- Remove deck plate before placing tips and pedestal onto deck layout

**Capacity:** Up to 5 racks of 96 cluster tubes (480 samples)

**Tips Utilized:** 8 - 1,000  $\mu$ L tips

**Pipette Used:** 8-channel (no individual wells)



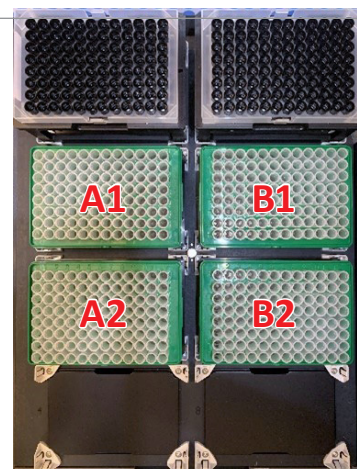
## Transfer-5 $\mu$ L-Cluster Tube to Cluster Tube

- 4 – Cluster Tube Rack Adapters
- 4 – Green Cluster Tube Racks w/Cluster Tubes
  - **Rack A1 & B1** – Containing 200  $\mu$ L sample or completed BHI regrowth sample (uncapped)
  - **Rack A2 & B2** – Pre-Filled Lysis Solution (uncapped)
- 2 – Rack of 50  $\mu$ L Framed Conductive Filter

**Capacity:** Up to 2 racks of 96 cluster tubes (192 samples)

**Tips Utilized:** 192 - 50  $\mu$ L tips

**Pipette Used:** 8-channel (no individual wells)



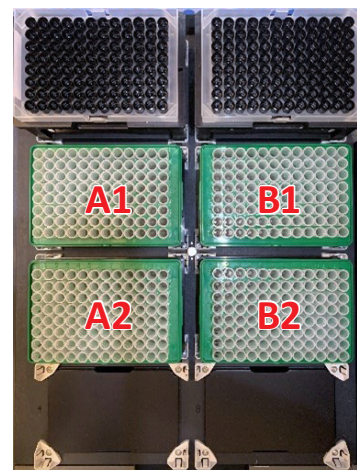
## Transfer-20 $\mu$ L-Cluster Tube to Cluster Tube

- 4 – Cluster Tube Rack Adapters
- 4 – Green Cluster Tube Racks w/Cluster Tubes
  - **Rack A1 & B1** – Containing 200  $\mu$ L sample or completed BHI regrowth sample (uncapped)
  - **Rack A2 & B2** – Pre-Filled Lysis Solution (uncapped)
- 2 – Rack of 50  $\mu$ L Framed Conductive Filter

**Capacity:** Up to 2 racks of 96 cluster tubes (96 samples)

**Tips Utilized:** 192 - 50  $\mu$ L tips

**Pipette Used:** 8-channel (no individual wells)



## Hydrate-30 $\mu$ L-Lysate to PCR

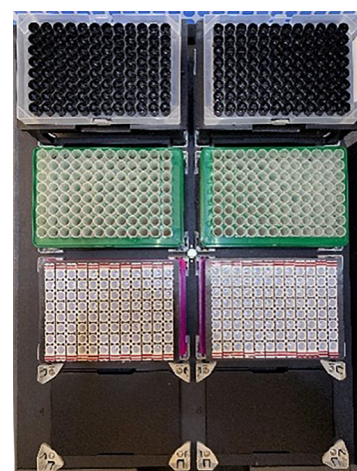
- 2 – PCR Coolers
  - Ensure coolers are kept in freezer prior to use
  - Uncap all PCR strips and immediately begin protocol
    - **Ensure black carrying tray is utilized with PCR strips**
- 2 – Green Cluster Tube Racks w/Cluster Tubes
  - Completed (heated and cooled) lysate samples (uncapped)
- 2 – Racks of 50  $\mu$ L Framed Conductive Filter

**Capacity:** Up to 2 racks of 96 cluster tubes (192 samples)

**Tips Utilized:** 192 - 50  $\mu$ L tips

**Pipette Used:** 8-channel and single-channel

- Single-channel (low throughput) should only be used when a single strip does not contain 8 wells. The user must complete the high throughput protocol first, then activate the single-channel protocol to finish the remainder of the rack (**Hydrate-30  $\mu$ L-Lysate to PCR SINGLE**).



## Hydrate-50 $\mu$ L-Lysate to PCR

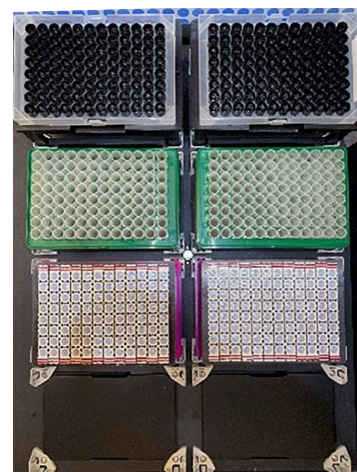
- 2 – PCR Coolers
  - Ensure coolers are kept in freezer prior to use
  - Uncap all PCR strips and immediately begin protocol
    - **Ensure black carrying tray is utilized with PCR strips**
- 2 – Green Cluster Tube Racks w/Cluster Tubes
  - Completed (heated and cooled) lysate samples (uncapped)
- 2 – Racks of 50  $\mu$ L Framed Conductive Filter

**Capacity:** Up to 2 racks of 96 cluster tubes (192 samples)

**Tips Utilized:** 192 - 50  $\mu$ L tips

**Pipette Used:** 8-channel and single-channel

- Single-channel (low throughput) should only be used when a single strip does not contain 8 wells. The user must complete the high throughput protocol first, then activate the single-channel protocol to finish the remainder of the rack (**Hydrate-50  $\mu$ L-Lysate to PCR SINGLE**).



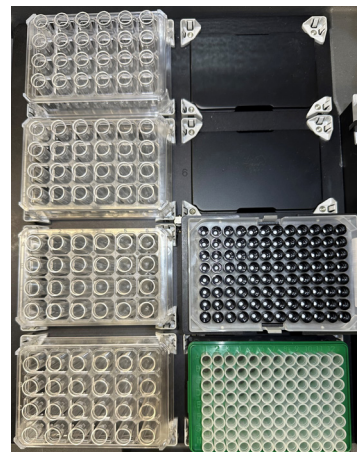
## Sample Tubes to Cluster Tube Rack

- 1 – Cluster Tube Rack Adapter
- 1 – Green Cluster Tube Racks w/Cluster Tubes
- 4 – Tube Racks each containing twenty-four 5 mL round bottom tubes (Falcon 352054 or similar) with at least 500  $\mu$ L enrichment sample
- 1 – Rack of 300  $\mu$ L Framed Conductive Filter Tips

**Capacity:** Up to 96 enrichment samples

**Tips Utilized:** 96 - 50  $\mu$ L tips

**Pipette Used:** Single channel(s) only



## Sample Tubes to Cluster Tube Rack - 5 $\mu$ L

- 1 – Cluster Tube Rack Adapter
- 1 – Green Cluster Tube Rack w/Cluster Tubes Pre-filled with 200  $\mu$ L lysis reagent (uncapped)
- 4 – Tube Racks each containing twenty-four 5 mL round bottom tubes (Falcon 352054 or similar) with at least 100  $\mu$ L enrichment sample
- 1 – Rack of 50  $\mu$ L Framed Conductive Filter Tips

**Capacity:** Up to 96 samples

**Tips Utilized:** 96 - 50  $\mu$ L tips

**Pipette Used:** Single channel(s) only



## Sample Tubes to Cluster Tube Rack - 10 $\mu$ L

- 1 – Cluster Tube Rack Adapter
- 1 – Green Cluster Tube Rack w/Cluster Tubes Pre-filled with 500  $\mu$ L BHI media (uncapped)
- 4 – Tube Racks each containing twenty-four 5 mL round bottom tubes (Falcon 352054 or similar) with at least 100  $\mu$ L enrichment sample
- 1 – Rack of 50  $\mu$ L Framed Conductive Filter Tips

**Capacity:** Up to 96 samples

**Tips Utilized:** 96 - 50  $\mu$ L tips

**Pipette Used:** Single channel(s) only



## Sample Tubes to Cluster Tube Rack - 20 $\mu$ L

- 1 – Cluster Tube Rack Adapter
- 1 – Green Cluster Tube Rack w/Cluster Tubes Pre-filled with 200  $\mu$ L Lysis Reagent (uncapped)
- 4 – Tube Racks each containing twenty-four 5 mL round bottom tubes (Falcon 352054 or similar) with at least 100  $\mu$ L enrichment sample
- 1 – Rack of 50  $\mu$ L Framed Conductive Filter Tips

**Capacity:** Up to 96 samples

**Tips Utilized:** 96 - 50  $\mu$ L tips

**Pipette Used:** Single channel(s) only

